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Article

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Evaluation of the prebiotic potential of arabinoxylans extracted from wheat distillers' dried grains with solubles (DDGS) and in-process samples

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Supplemental Table S1: AXOS/XOS hydrolysates assimilation in pH-controlled batch cultures after inoculation at 0 h, 4 h and 8 h after fermentation. Values are expressed as data percentage of total peak area (nC*min) relative to time 0.

		Time 0 h	Time 4 h	Time 8 h
DDGS-Xyl	Xylose	47.07 (13.57)	35.66 (4.72)	7.38 (7.70)
	Xylobiose	1.13 (0.08)	2.67 (0.93)	0.84 (0.96)
	Xylotriose	24.21 (20.47)	7.42 (7.27)	1.92 (2.61)
	Xylo-tetraose	3.69 (3.06)	10.00 (4.87)	2.45 (0.82)
	Xylo-pentaose	3.50 (2.12)	3.99 (3.29)	2.10 (1.06)
	Xylo-hexaose	3.79 (3.98)	6.24 (7.13)	3.91 (4.35)
	XA3XX	2.35 (2.00)	4.60 (3.28)	1.23 (1.64)
DDGS-Xyl+FAE	Xylose	33.50 (4.70)	16.80 (10.81)	0.25 (0.43)
	Xylobiose	16.80 (1.24)	14.32 (0.43)	2.50 (4.33)
	Xylotriose	14.93 (7.83)	12.20 (4.74)	0.06 (0.10)
	Xylo-tetraose	9.75 (0.99)	7.65 (1.69)	1.47 (2.55)
	Xylo-pentaose	1.35 (0.21)	1.21 (2.20)	0.60 (1.03)
	Xylo-hexaose	2.95 (2.70)	0.68 (0.79)	0.60 (1.03)
	XA3XX	5.39 (0.71)	0.77 (0.75)	*ND
WS-Xyl	Xylose	29.75 (8.10)	32.12 (10.81))	2.24 (2.30)
	Xylobiose	3.56 (1.32)	5.08 (0.43)	2.98 (2.64)
	Xylotriose	12.78 (8.32)	14.09 (4.74)	7.26 (8.47)
	Xylo-tetraose	13.25 (1.85)	14.75 (1.69)	6.61 (6.68)
	Xylo-pentaose	7.58 (0.57)	8.10 (2.20)	5.36 (4.66)
	Xylo-hexaose	4.20 (0.96)	4.27(0.79)	4.46 (4.34)
	XA3XX	4.49 (0.92)	3.82 (2.32)	4.95 (4.80)
WS-Xyl+FAE	Xylose	31.16 (11.51)	40.17 (11.85)	6.79 (8.46)
	Xylobiose	3.09 (1.00)	4.92 (1.40)	3.85 (3.34)
	Xylotriose	5.61 (2.89)	8.16 (3.55)	2.03 (1.82)
	Xylo-tetraose	10.58 (0.59)	13.87 (4.37)	9.20 (13.64)
	Xylo-pentaose	7.39 (1.27)	5.49 (2.17)	5.48 (5.27)
	Xylo-hexaose	4.59 (1.33)	4.31 (0.83)	4.13 (3.62)
	XA3XX	3.03 (0.28)	2.10 (1.83)	2.44 (2.49)

*ND. Non-detected. Standard deviation is shown in parentheses ($n = 3$). Oligosaccharides obtained from DDGS and WS were totally depleted at 24 h of fermentation

Supplemental Table S2: Mean bacterial populations in pH-controlled batch cultures at 0, 4, 8 and 24 h

Probe	Time (h)	Bacterial population (\log_{10} cells ml^{-1} batch culture fluid) with substrate:					
		Negative control	FOS	DDGS-Xyl	DDGS -Xyl+ FAE	WS-Xyl	WS-Xyl + FAE
BIF 164	0	6.85 (0.25)a	6.88 (0.25)a	7.14 (0.03)a	7.00 (0.17)a	6.97 (0.10)a	6.95 (0.18)a
	4	6.91 (0.30)a	7.54 (0.36)a	8.28 (0.42)a*	8.01 (1.06)a	7.65 (0.51)a	7.70 (0.45)a*
	8	6.77 (0.16)a	7.63 (0.49)ab	8.47 (0.54)b*	8.27 (0.79)ab	7.16 (0.70)ab	7.87 (0.16)ab*
	24	6.76 (0.18)a	8.20 (0.33)b*	8.70 (0.17)b*	8.33 (0.82)b*	8.66 (0.77)b*	8.32 (0.18)b*
LAB 158	0	6.16 (0.78)a	5.92 (0.92)a	6.27 (0.59)a	5.99 (0.63)a	5.85 (0.65)a	5.91 (0.89)a
	4	5.68 (0.46)a	6.10 (0.70)a	6.14 (0.08)a	5.95 (0.35)a	5.99 (0.61)a	5.45 (0.12)a
	8	5.76 (0.11)a	5.65 (0.70)a	6.07 (0.33)a	6.13 (0.36)a	6.24 (1.03)a	5.95 (0.44)a
	24	5.77 (0.23)a	6.15 (0.35)a	6.86 (0.29)a	6.47 (0.38)a	7.30 (0.38)a	6.98 (0.91)a
BAC 303	0	6.87 (0.07)a	6.54 (0.80)a	6.79 (0.37)a	6.58 (0.43)a	6.49 (0.25)a	6.42 (0.31)a
	4	6.58 (0.21)a	7.07 (0.50)a	6.78 (0.90)a	6.56 (1.33)a	6.78 (0.36)a	6.77 (0.52)a
	8	7.07 (0.63)a	6.58 (0.78)a	6.80 (0.73)a	6.48 (1.58)a	6.50 (1.14)a	6.29 (0.43)a*
	24	6.75 (0.25)a	6.12 (0.49)a	8.07 (0.36)a	7.16 (1.20)a	8.22 (0.89)a	7.78 (0.45)a*
EREC 482	0	8.08 (0.35)a	8.09 (0.26)a	8.28 (0.10)a	8.08 (0.11)a	8.14 (0.17)a	8.12 (0.20)a
	4	7.79 (0.00)a	8.25 (0.18)a	8.36 (0.17)a	8.14 (0.75)a	8.38 (0.29)a	8.27 (0.32)a
	8	7.91 (0.23)a	7.33 (0.65)a	8.40 (0.20)a	8.47 (0.37)a	8.02 (0.99)a	8.69 (0.71)a
	24	7.74 (0.23)a	7.56 (0.35)a	8.64 (0.62)a	8.62 (0.50)a	8.34 (0.64)a	7.94 (0.90)a
RREC 584	0	6.84 (0.31)a	6.83 (0.25)a	7.16 (0.42)a	6.89 (0.31)a	6.98 (0.42)a	6.95 (0.55)a
	4	6.16 (1.13)a	6.82 (0.52)a	6.49 (0.63)a	6.48 (0.75)a	6.90 (1.03)a	6.77 (0.88)a
	8	6.60 (0.24)a	6.18 (0.97)a	6.76 (0.73)a	6.90 (1.16)a	6.69 (1.14)a	6.93 (1.54)a
	24	6.26 (0.17)a	5.74 (0.49)a	7.30 (1.10)a	6.96 (0.91)a	7.16 (1.17)a	6.89 (1.49)a
ATO 291	0	6.18 (1.52)a	5.92 (1.20)a	6.15 (1.01)a	5.76 (1.27)a	5.83 (1.23)a	5.88 (1.26)a
	4	6.70 (1.18)a	6.63 (0.59)a	6.60 (1.43)a	6.79 (0.55)a	6.64 (1.40)a	6.49 (1.39)a
	8	6.77 (0.79)a	6.02 (1.26)a	6.81 (1.18)a	6.35 (1.31)a	6.10 (1.45)a	6.74 (1.44)a
	24	6.52 (1.13)a	6.08 (1.44)a	6.99 (1.23)a	6.87 (0.77)a	6.55 (1.29)a	6.70 (1.25)a
PRO 853	0	6.66 (0.04)a	6.47 (0.75)a	6.75 (0.14)a	6.60 (0.49)a	6.58 (0.30)a	6.36 (0.69)a
	4	6.28 (0.68)a	7.02 (0.86)a	6.83 (1.06)a	6.50(1.21)a	6.49 (0.75)a	6.78 (0.54)a
	8	7.18 (0.75)a	6.70 (0.80)a	7.10 (0.76)a	6.81 (1.29)a	6.69 (0.89)a	7.00 (0.39)a
	24	6.62 (0.16)a	6.55 (0.27)a	7.99 (0.37)a	7.28 (0.90)a	8.16 (0.83)a	7.83 (0.45)a*
FPRAUTZ 655	0	7.58 (0.48)a	7.64 (0.34)a	7.88 (0.15)a	7.68 (0.15)a	7.75 (0.18)a	7.70 (0.32)a
	4	7.18 (0.72)a	7.71 (0.33)a	7.57 (0.54)a	7.22 (0.45)a	7.88 (0.61)a	7.64 (0.70)a
	8	7.38 (0.11)a	6.77 (0.90)*	6.92 (1.16)a	6.95 (0.71)a	7.34 (0.84)a	7.64 (1.06)a
	24	6.89 (0.25)ab	6.45 (0.15)a*	7.99 (0.36)b	7.47 (0.17)ab	7.93 (0.62)b	7.50 (0.64)ab
DSV 687	0	7.38 (0.23)a	7.37 (0.12)a	7.65 (0.15)a	7.42 (0.10)a	7.50 (0.10)a	7.53 (0.20)a
	4	7.02 (0.81)a	7.28 (0.33)a	7.27 (0.91)a	6.77 (0.61)a	7.50 (0.60)a	7.19 (0.74)a
	8	6.76 (0.19)a	6.33 (1.10)a	6.31 (0.98)a	6.12 (1.09)a	6.88 (0.53)a	6.51 (1.31)a
	24	6.45 (0.57)a	6.19 (0.71)a	7.11 (0.49)a	7.01 (0.57)a	7.05 (0.67)a	6.80 (1.22)a
CHIS 150	0	6.16 (0.53)a	6.17 (0.37)a	6.42 (0.18)a	6.30 (0.28)a	6.30 (0.38)a	6.35 (0.60)a
	4	6.15 (0.96)a	6.14 (0.41)a	6.37 (0.74)a	6.05 (1.03)a	6.26 (0.90)a	5.78 (1.05)a
	8	6.24 (0.20)a	5.70 (0.62)a	6.17 (0.48)a	6.42 (0.81)a	5.73 (0.55)a	6.15 (0.55)a
	24	5.84 (0.05)a	6.23 (0.63)a	6.77 (0.11)a	6.78 (0.34)a	6.83 (0.09)a	6.58 (0.60)a

Standard deviation is shown in parentheses (n = 3). 2 way ANOVA with Tukey's post hoc tests were used for the statistical analysis. Significant differences ($P < 0.05$) among treatment at the same time point are indicated with different letters. (*) Significant different from 0 h value, $P < 0.05$).

Supplemental Table S3: Mean SCFAs and lactic acid concentrations in pH-controlled batch cultures at 0, 4, 8, and 24 h.

	Lactate	Formic	Acetic	Propionic	Butyric	Total SCFAa
No treatment						
0 h	0.32 (0.47)a	0.06 (0.06)a	7.34 (3.74)a	2.61 (3.98)a	2.58 (1.13)a	12.91 (5.66)a
4 h	1.08 (1.49)a	0.03 (0.06)a	7.30 (6.27)a	17.67 (27.66)a	1.83 (1.16)a	27.92 (28.86)a
8 h	0.22 (0.33)a	0.06 (0.11)a	10.23 (8.46)a	10.55 (14.26)a	1.31 (1.64)a	22.38 (17.77)a
24 h	0.03 (0.06)a	0.05 (0.09)a	12.90 (10.90)a	3.03 (1.27)a	1.55 (2.21)a	17.56 (12.27)a
FOS						
0 h	0.65 (0.32)a	0.37 (0.23)a	8.13 (3.81)a	4.71 (7.87)a	1.79 (0.80)a	15.64 (6.39)a
4 h	3.50 (1.21)a	1.17 (1.77)a	18.86 (15.21)a	4.09 (3.97)a	1.06 (1.43)a	28.67 (15.44)a
8 h	19.20 (8.94)b*	10.97 (12.62)a	43.11 (28.10)ab	15.60 (10.61)a	3.14 (3.94)a	92.02 (43.02)ab*
24 h	4.75 (8.01)a	8.92 (10.79)a	65.26 (20.41)b*	22.67 (15.13)a	2.61 (3.57)a	104.21 (35.04)bc*
DGGS-Xyl						
0 h	1.16 (0.79)a	0.57 (0.64)a	10.21 (5.60)a	2.70 (4.51)a	1.76 (1.85)a	16.40 (5.72)a
4 h	2.83 (2.50)a	9.52 (5.77)a	32.98 (13.92)a	13.53 (16.87)a	1.57 (0.86)a	60.44 (21.63)a
8 h	5.79 (2.33)a*	9.53 (6.41)a	51.98 (8.04)ab**	14.56 (5.04)a	3.17 (2.80)a	85.03 (10.18)ab**
24 h	0.14 (0.23)a±	3.78 (6.55)a	58.99 (13.80)b**	18.42 (2.44)ab	10.38 (6.63)a	91.70 (26.10)bc**
DGGS-Xyl + FAE						
0 h	1.44 (1.37)a	1.77 (1.55)a	18.01 (5.61)a	4.46 (2.07)a	2.49 (2.28)a	28.15 (6.00)a
4 h	4.03 (2.38)a	8.41 (7.17)a	31.21 (19.02)a	4.37 (3.44)a	2.24 (3.18)a	50.25 (33.89)a
8 h	8.31 (2.90)ab*	8.08 (1.27)a	49.04 (21.30)ab	11.45 (9.91)a	5.80 (3.56)a	82.69 (35.18)ab
24 h	0.06 (0.10)a±	0.65 (1.12)a	55.30 (4.51)b	14.12 (4.24)ab	5.45 (5.05)a	75.58 (12.37)ab
WS-Xyl						
0	1.97 (0.55)a	0.89 (0.71)a	10.24 (1.35)a	4.03 (6.73)a	1.91 (0.47)a	19.04 (8.36)a
4	5.90 (3.34)a	3.91 (1.16)a	17.73 (6.34)a	23.51 (20.14)a	0.97 (0.66)a	52.02 (19.98)a
8	3.07 (3.74)a	13.59 (8.41)a	63.86 (2.55)b***	22.51 (11.65)a	6.45 (10.22)a	109.48 (16.97)b**
24	3.40 (5.72)a	4.43 (6.83)a	87.88 (3.12)b***	44.37 (20.75)b	7.37 (8.82)a	147.45 (28.53)c***
WS-Xyl + FAE						
0	0.60 (0.26)a	0.17 (0.29)a	7.34 (2.57)a	6.35 (5.53)a	1.47 (0.70)a	15.93 (4.11)a
4	3.12 (0.60)a	4.68 (0.78)a	23.59 (2.85)a	13.83 (10.98)a	0.97 (0.60)a	46.18 (9.78)a*
8	1.91 (1.49)a*	9.48 (6.53)a	58.52 (12.62)b***	15.92 (7.23)a	1.16 (0.72)a	86.99 (11.26)ab**
24	0.24 (0.06)a±	3.82 (6.56)a	58.52 (11.84)b***	19.76 (4.35)ab	3.37 (1.15)a	85.71 (9.54)bc**

Standard deviation is shown in parentheses ($n = 3$). 2-way ANOVA with Tukey's post hoc tests were used for the statistical analysis. Significant differences ($P < 0.05$) among treatment at the same time point are indicated with different letters. (*) Significant difference from 0 h value, $P < 0.05$.

Supplemental Table S4: Mean bacterial populations in pH –controlled mini batch cultures using FOS, XOS or AXOS/XOS DP ≥ 3 as substrates at 0,8 and 24 h of fermentation.

Probe	Time (h)	Bacterial population (\log_{10} cells ml^{-1} batch culture fluid) with substrate			
		Negative control	FOS	XOS	AXOS ≥ 3
EUB	0	7.77 (0.33)	7.68 (0.24)	7.67 (0.26)	7.88 (0.04)
	8	7.53 (0.59)	8.43 (0.47)	8.41 (0.36)	8.03(0.46)
	24	7.65 (0.42)	8.28 (0.33)	8.39 (0.33)	8.18 (0.11)
BIF	0	7.06 (0.45)	6.79 (0.27)	6.80 (0.47)	7.04 (0.42)
	8	6.88 (0.67)a	8.29 (0.49)b	8.29 (0.39)b	7.61 (0.87)a
	24	6.72 (0.35)a	8.14 (0.42)b	8.28 (0.33)b	7.91 (0.30)ab
LAB	0	5.90 (0.48)	5.79 (0.39)	5.73 (0.32)	5.90 (0.34)
	8	5.93 (0.65)	6.23 (0.50)	5.88 (0.66)	5.81 (0.29)
	24	6.02 (0.32)	5.24 (0.29)	5.50 (0.54)	6.08(0.10)
BAC	0	6.20 (0.55)	6.16 (0.56)	6.12 (0.45)	6.28 (0.29)
	8	6.21 (0.47)	6.56 (0.21)	6.33 (0.07)	6.36 (0.09)
	24	6.29 (0.28)	5.80 (0.24)	5.79 (0.59)	6.64 (0.07)
EREC	0	7.27 (0.26)	7.20 (0.18)	7.17 (0.20)	7.34 (0.08)
	8	6.70 (0.54)	7.52 (0.51)	7.38 (0.44)	7.28 (0.15)
	24	6.59 (0.4)	7.48 (0.23)	7.48 (0.41)	6.88 (0.53)
RREC	0	6.42 (0.41)	6.44 (0.34)	6.33 (0.45)	6.60 (0.33)
	8	5.69 (0.45)	6.41 (0.62)	5.98 (0.36)	6.36 (0.01)
	24	6.01 (0.3)	5.78 (0.63)	5.52 (0.46)	6.26 (0.26)
ATO	0	5.63 (0.92)	5.43 (1.00)	5.62 (0.60)	5.92 (0.63)
	8	5.57 (0.92)	6.06 (0.81)	5.54 (0.80)	5.91 (0.14)
	24	6.29 (0.7)	5.38 (0.33)	5.76 (0.75)	5.96 (0.33)
PRO	0	6.22 (0.54)	6.22 (0.57)	6.15 (0.47)	6.32 (0.28)
	8	6.36 (0.35)	6.58 (0.13)	6.66 (0.26)	6.47 (0.21)
	24	6.30 (0.3)	5.93 (0.10)	5.98 (0.40)	6.76 (0.17)
FPRAUTZ	0	6.92 (0.19)	6.96 (0.22)	6.93 (0.20)	7.05 (0.03)
	8	6.63 (0.57)	6.85 (0.41)	6.92 (0.29)	6.78 (0.13)
	24	6.23 (0.21)	6.47 (0.68)	6.34 (0.56)	6.50 (0.53)
DSV	0	6.61 (0.22)	6.77 (0.33)	6.62 (0.30)	6.85 (0.11)
	8	6.33 (0.55)	6.64 (0.41)	6.39 (0.29)	6.49 (0.16)
	24	6.14 (0.43)	6.04 (0.54)	5.96 (0.34)	6.37 (0.39)
CHIS	0	6.14 (0.24)	5.96 (0.15)	5.98 (0.17)	6.11 (0.02)
	8	5.73 (0.29)	6.36 (0.25)	6.15 (0.42)	6.31 (0.73)
	24	6.19 (0.33)	5.48 (0.29)	5.51 (0.28)	6.25 (0.20)

Standard deviation is shown in parentheses (n = 3). 2 way ANOVA with Tukey's post hoc tests were used for the statistical analysis. Significant differences ($P < 0.05$) among treatment at the same time point are indicated with different letters. (*) Significant different from 0 h value, $P < 0.05$).